

Abstract No. Kuzm0303

Molecular Structure of Lipopolysaccharides in Thin Films at the Air-water Interface

I. Kuzmenko (HHMI, UC Berkeley), B. Ocko (BNL), C. Bertozzi (HHMI, UC Berkeley)
Beamline(s): X22B

Introduction: Lipopolysaccharides constitute outer membranes of Gram-negative bacteria and mediate a host of immune responses in mammals potentially causing fever, shock and even death. These molecules are composed of oligosaccharide core (two sugar units) with several fatty-acid tails attached to it, which makes the whole molecule amphiphilic. Lipid A, which is one of the simplest representatives of the family, is depicted in Fig.1a. Structural motifs of lipopolysaccharides were previously studied in thin 3D crystals (AFM, powder diffraction, or electron microscopy).[1,2] The knowledge of the molecular packing of lipopolysaccharide films at the air-water interface, may provide a better insight into the supramolecular structure of the lipids within living membranes and into their function.

Methods and Materials: Grazing incidence X-ray diffraction (GIXD) and X-ray reflectivity (XR) were employed to study monolayers of lipid A on water. Lipid A, diphosphoryl and monophosphoryl were purchased from Sigma and used without further purification (catalog numbers L-0774, L-5399, L-5538). Compounds were dissolved in chloroform and spread drop-wise on the liquid surface. Surface pressure - molecular area (π -A) isotherms were recorded prior to X-ray measurements to check the amphiphilic properties of the compounds.

Results: According to the (π -A) isotherms all of the compounds form monolayers both on water and on 0.1M aqueous solution of calcium chloride (Fig.1b). Grazing incidence X-ray diffraction pattern contains a single peak at $q_{xy} = 1.46 \text{ \AA}^{-1}$ which corresponds to a hexagonal packing of the untilted hydrocarbon chains. (Fig.1c). According to the structural factor calculation of the Bragg rod (Fig.1d), sugar moieties do not contribute to the diffraction pattern and appear to be disordered. The presence of calcium ions in the solution does not play any noticeable role in the structural organization of the film, as was suggested in other studies.[1] Complementary X-ray reflectivity measurements support the monolayer formation in all cases.

Conclusions: Present studies of several lipopolysaccharides at the air-liquid interface suggest that within biological membranes the hydrocarbon chains of the lipid adopt hexagonal packing with chain axis perpendicular to the interface whereas the 2D order is not maintained for the rest of the molecule.

Acknowledgments: Highlight this text and type over it with any acknowledgments you may have; otherwise, delete this line

- References**
- [1] Y. Wang and R. Hollingsworth, *Biochemistry*, **1996**, 35, 5647.
 - [2] N. Kato, T. Sugiyama *et al*, *Molecular Microbiology*, **2000**, 36, 796.

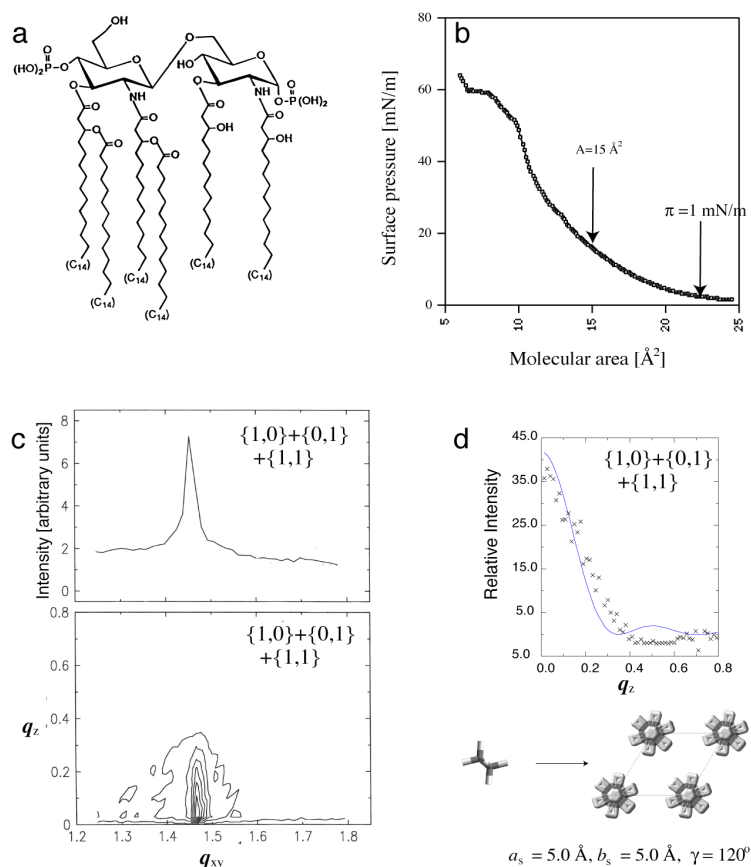


Figure 1. (a) Molecular structure of lipid A such as found in *E. coli* strains. (b) Typical surface pressure-molecular area (π -A) isotherms for lipid A, diphosphoryl on water and on 0.1M calcium chloride solution. Arrows mark points at the isotherm where GIXD and XR measurements were performed. (c) GIXD spectrum for lipid A, diphosphoryl on water at $\pi=20\text{mN/m}$ (d) Bragg rod calculated (solid line) vs. observed (crosses) data and the corresponding molecular model used in the structure factor computation: hexagonally packed C₁₄ hydrocarbon chains (rotator phase).